X-RAY MICROSCOPY BY PHASE-RETRIEVAL METHODS AT THE ADVANCED LIGHT SOURCE

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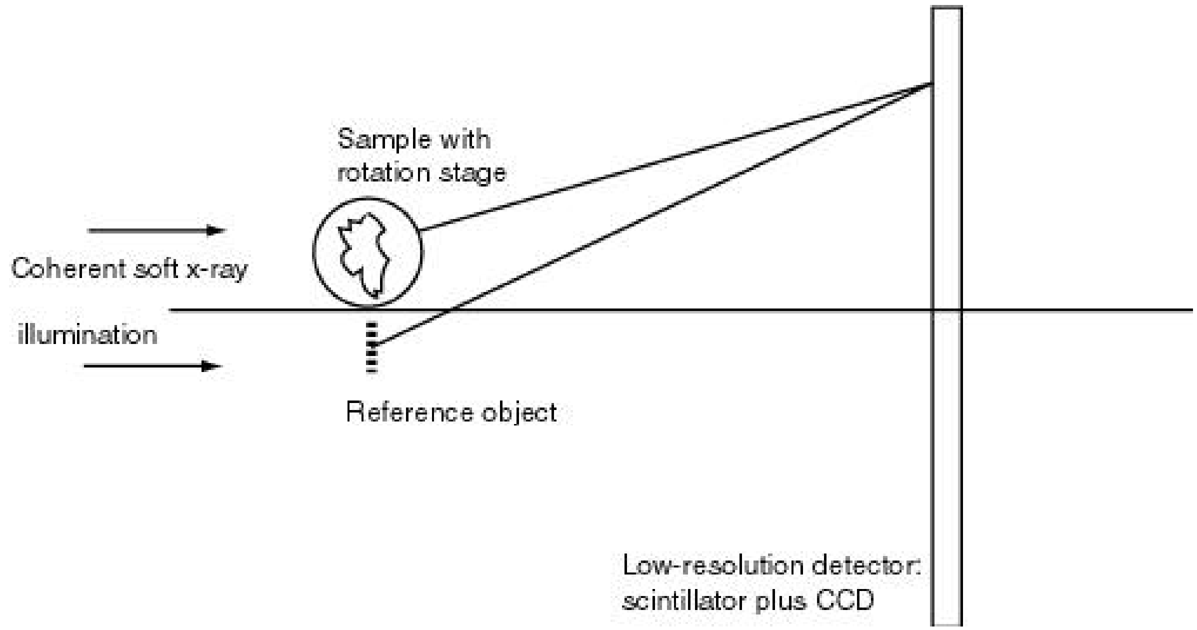
3-D DIFFRACTIVE IMAGING: MY STARTING POINT



- This is Fourier-transform holography with a complicated reference object
- Reference object could be known by independent measurement which would allow recovery of an image of the sample by holography
- Even if it were not known, the whole (double) object can be recovered by phase-retrieval algorithms which work particularly well for double objects - thereafter the reference object would be known
- Thus we have, in principle, a pathway to 3-D tomography if the reference object is kept fixed and the sample is rotated - imaginable but hard to do

IMAGING BY DIFFRACTION FROM A SINGLE OBJECT

- Experiment is pure diffraction by a single object (i. e. no holographic reference object)
- Original idea by Sayre 1980
- First realization - Miao, Charalambous, Kirz, Sayre 1999
- Depends on the power of the phasing algorithms developed by Fienup, Fiddy et al in the 1980's and now widely used in science and technology
- Although Miao et al have been very successful, surprisingly few experiments by other groups in *microscopy* of any sort have been done
- We have made a start with experiments using visible-light and electron microscopes and have produced successful reconstructions in both cases
- We learned from these experiments and from the literature that reconstructing a general complex object of unknown support (the most interesting type) in 2-D is much harder than reconstructing a real object
- We believe that this does not matter since one can either (a) use HXR where objects tend to be real or (b) move to 3-D which we believe to be easier

OVER- AND UNDER-DETERMINED PHASE PROBLEMS

R. Millane 1990, 1996, Sayre 1952

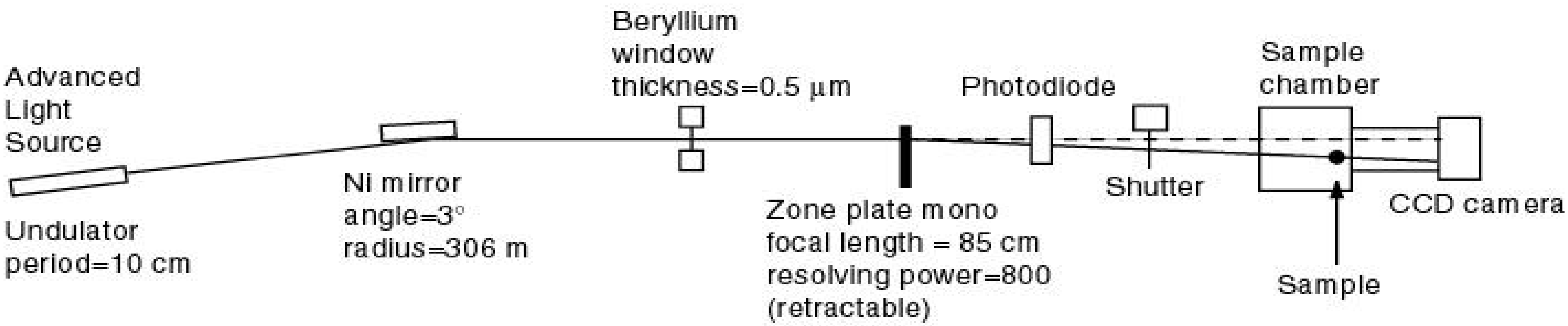
ITEM	VALUE
Width of object function (f)	a
Nyquist interval for Fourier amplitude (F)	1/a
Width of autocorrelation of f	2a
Nyquist interval for Fourier intensity $\|F\|^2$	1/2a
Bragg interval	1/a

- The Nyquist density of samples is twice as high for $|F|^2$ as it is for F
- Bragg sampling of $|F|^2$ (using reciprocal lattice points) is a factor two <u>undersampled</u> according to Nyquist, Shannon, Kotel'nikov etc
- The correct (Nyquist) sampling rate for $|F|^2$ is two times <u>oversampled</u> for F - this leads to blank spaces around the object upon passing from F to f (the inverse of Fourier interplolation)

# Dims	**Crystallography**		**Optics**	
	Unique?	**Over-determined?**	**Unique?**	**Over-determined?**
1			No	Under
2			Maybe	Not either
3	No	Under by 4	Yes	Over by 2

Conclusion:
Phase retrieval should be easier in 3-D than 2-D - nevertheless we think we should practice on 2-D first before progressing to 3-D

ALS BEAM LINE 9.0.1 COHERENT OPTICS

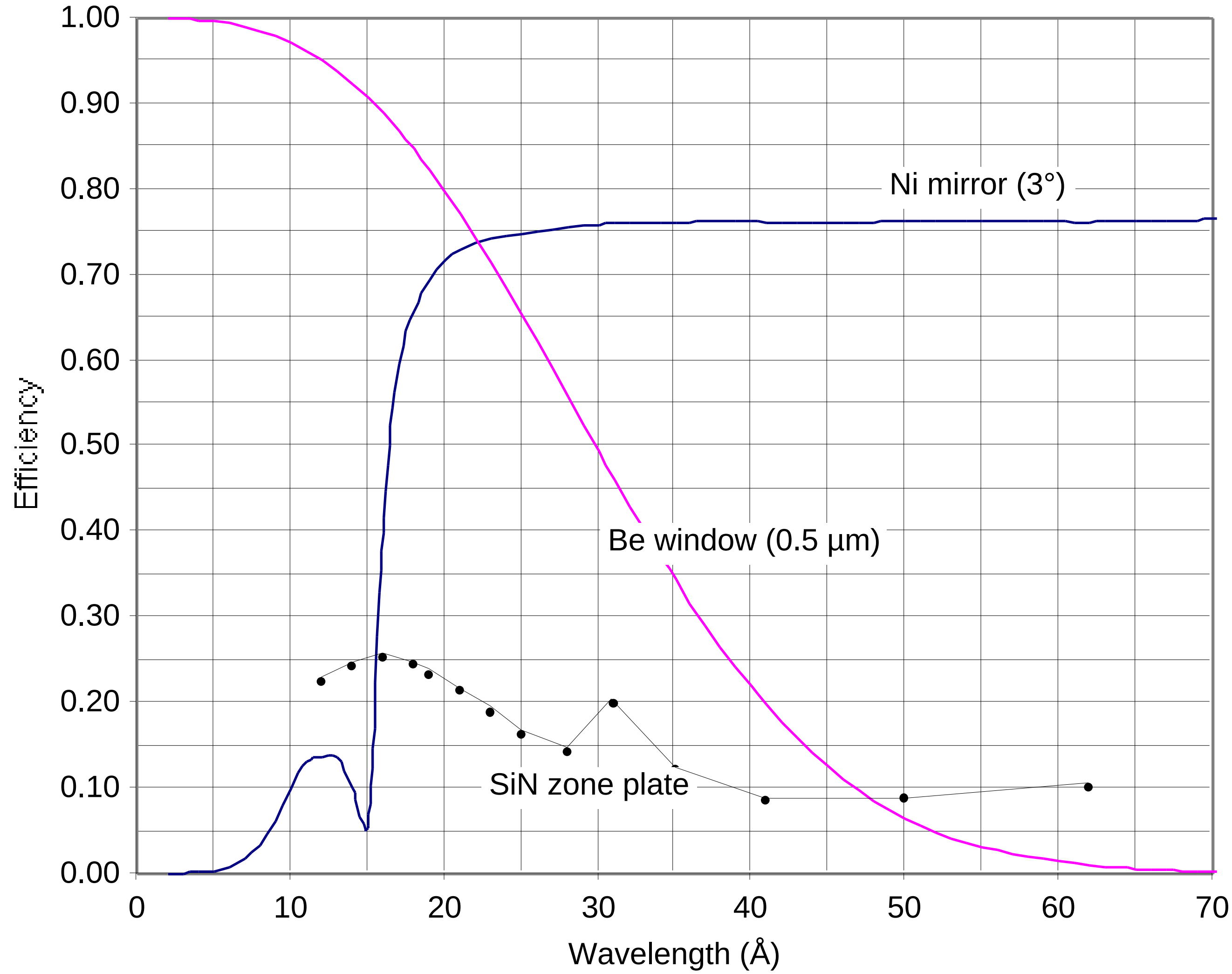


BEAM LINE PLAN VIEW NOT TO SCALE

NOTES:

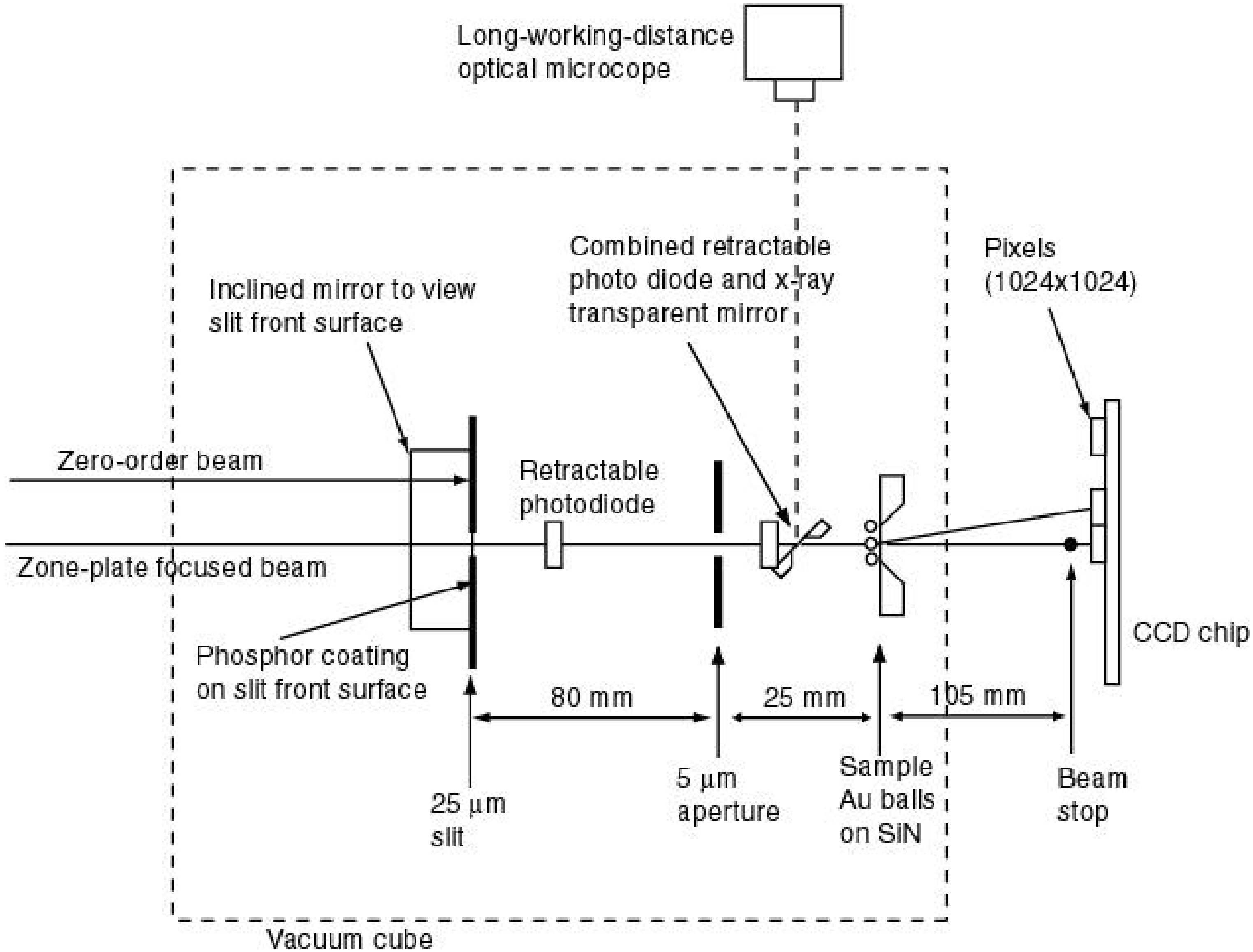
- Experiments are done at 588 eV in undulator 3rd harmonic
- Be window and zone-plate monochromator both 0.8 mm in size are designed to withstand pink (once reflected) beam
- Diffractive elements of the zone plate mono (Charalambous) made of silicon nitride coated on both sides with aluminium for better mechanical stability and heat removal

BEAM LINE EFFICIENCY FACTORS



M. R. Howells, Advanced Light Source

SOFT-X-RAY DIFFRACTION EXPERIMENT



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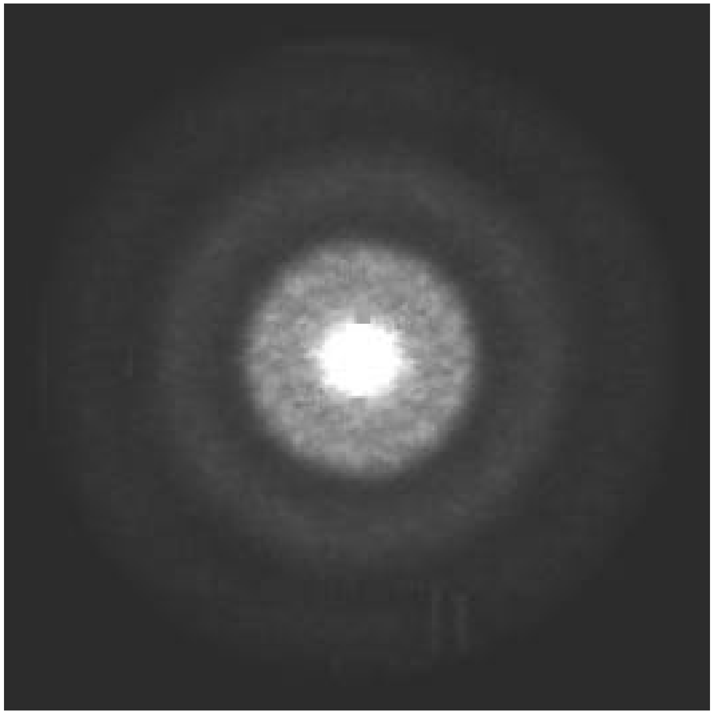
FIRST GOOD DATA ON GOLD-BALL SAMPLE



- 50 nm gold balls, effectively a real object so they give a symmetrical pattern (Friedel’s law)
- 500 µm SiN window
- This data has problems:
  - Missing cone in the center
  - Errors due to merging data sets - about 95% obeys Friedel's law - for crystallography data it would be about 99%
  - Object is not isolated (of finite support) which is needed for use of Fienup etc

- Fienup retrieval failed - not sure which "problem" is responsible - missing cone does not cause failures in simulations
- Decided to try reconstruction without knowing the missing cone
- Approaches: (1) direct methods (2) small windows

APPLICATION OF CRYSTALLOGRAPHIC DIRECT METHODS TO IMAGING

Giacovazzo 1989, Wolfson, 1995, 1997, Sayre 1952

Basic ideas:

- Requirements are:
  - atomicity
  - atomic resolution data
  - many measured beams per atom
- Nonrequirements:
  - missing cone no problem,
  - lack of isolation no problem
- We treat the gold balls as atoms
- Treat each detector pixel as a measured beam

Outcome:

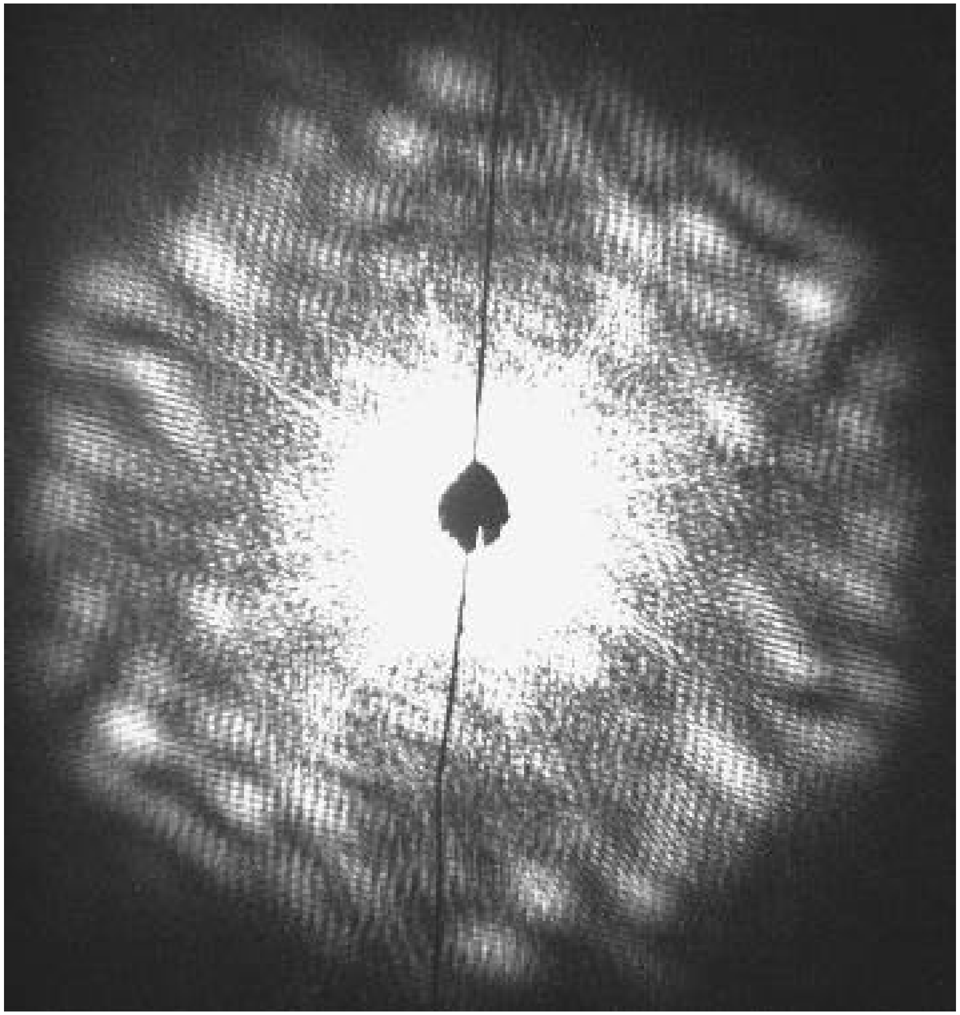
- Although we have reconstructed images with R factors as low as 0.22 using the SIR 97 program we do not yet have a reconstruction that we believe in
- This is the first time according to Giacovazzo that direct methods have been used on a non-periodic object and it is considered exciting in the direct-methods community

SMALL WINDOWS

G. Denbeaux

- Small SiN windows of around 2×2 μm have solved many of our problems and within the last month we have acquired much better data
- They reduce the size and strength of the central bright spot
- We have actually taken data with the beam stop removed (no missing cone) but have not yet analyzed it
- They produce much less stray X-ray signal due to edge scattering - the edge is too short to have much scattering power
- Their diffraction pattern is visible outside the beam-stop from which the dimensions of the window can be found.
- They provide sample isolation inside a known and tight support
- Through the window is the only path for stray light in a well-baffled system so stray light is reduced in proportion to window size
- Clarifies the issue of how large an area needs to be coherently illuminated or indeed be illuminated at all

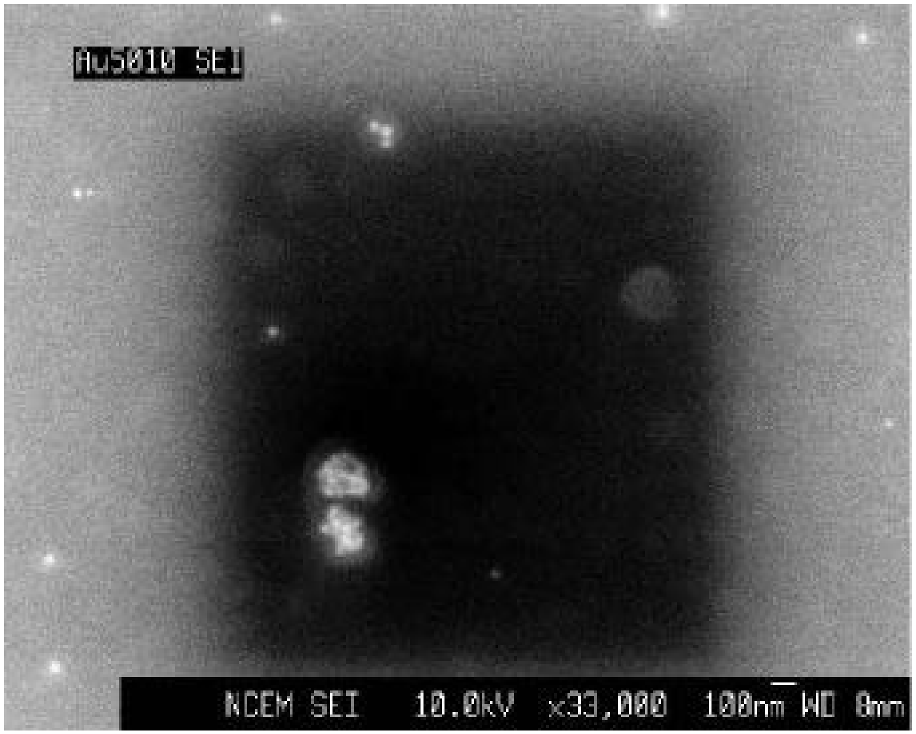
IMPROVED DATA FROM SMALL-WINDOW SAMPLE



- 30 nm gold balls 588 eV
- 1.77x1.77 µm window from window diffraction pattern

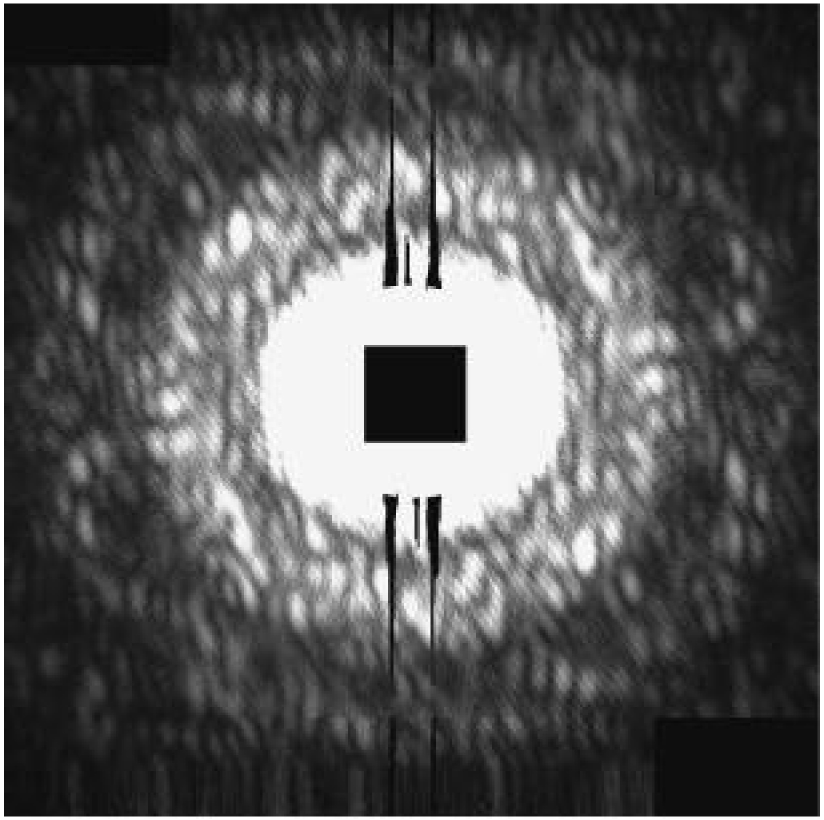
- Smaller beam stop
- No merging of data sets
- Good statistics: sum of many short runs
- No SEM image or reconstruction yet
- Features:
  - fringes - show separated parts and understandable phase encoding
  - Hexagonal shape - 2-D packing of balls
  - Friedel symmetry

SEM PICTURE OF A GOLD-BALL SAMPLE

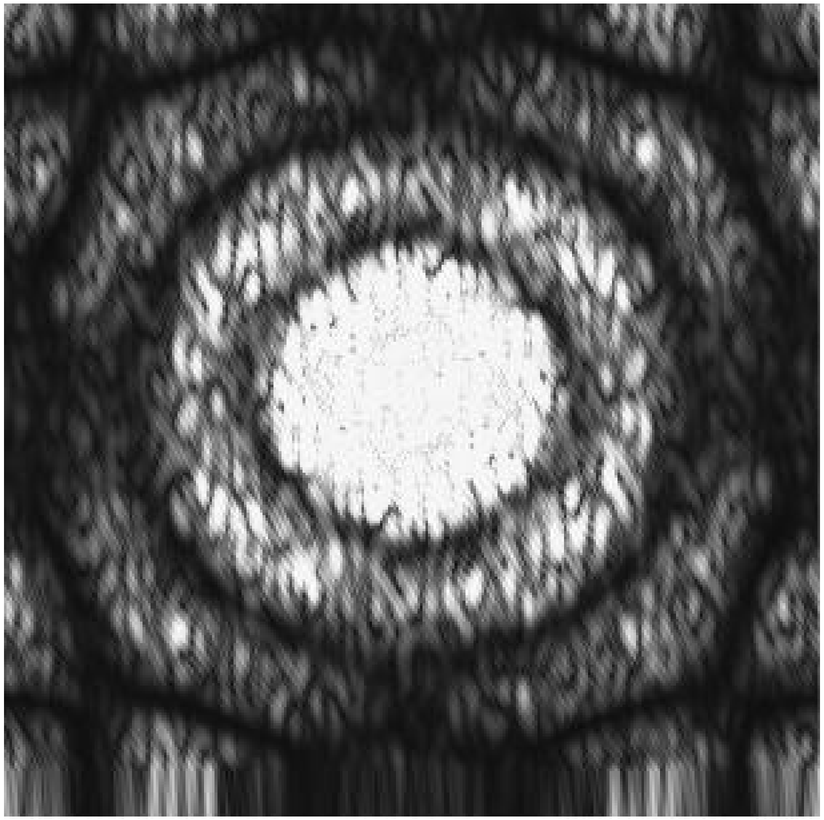


- SEM image of sample that was reconstructed in the last two weeks
- 50 nm gold balls
- “Window”=2x2 micron roughly

DIFFRACTION PATTERNS

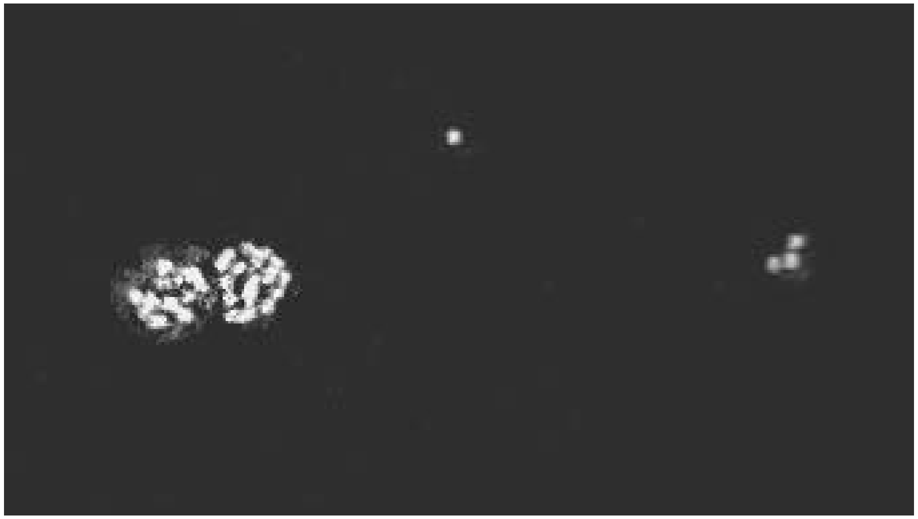


50 nm gold balls, 588eV- measured pattern



Same pattern calculated from the SEM image

OBJECT RECONSTRUCTION



HiO reconstruction

Positivity applied to real and imaginary parts of the transparency function

No use of ballicity or binary character or pure gold character

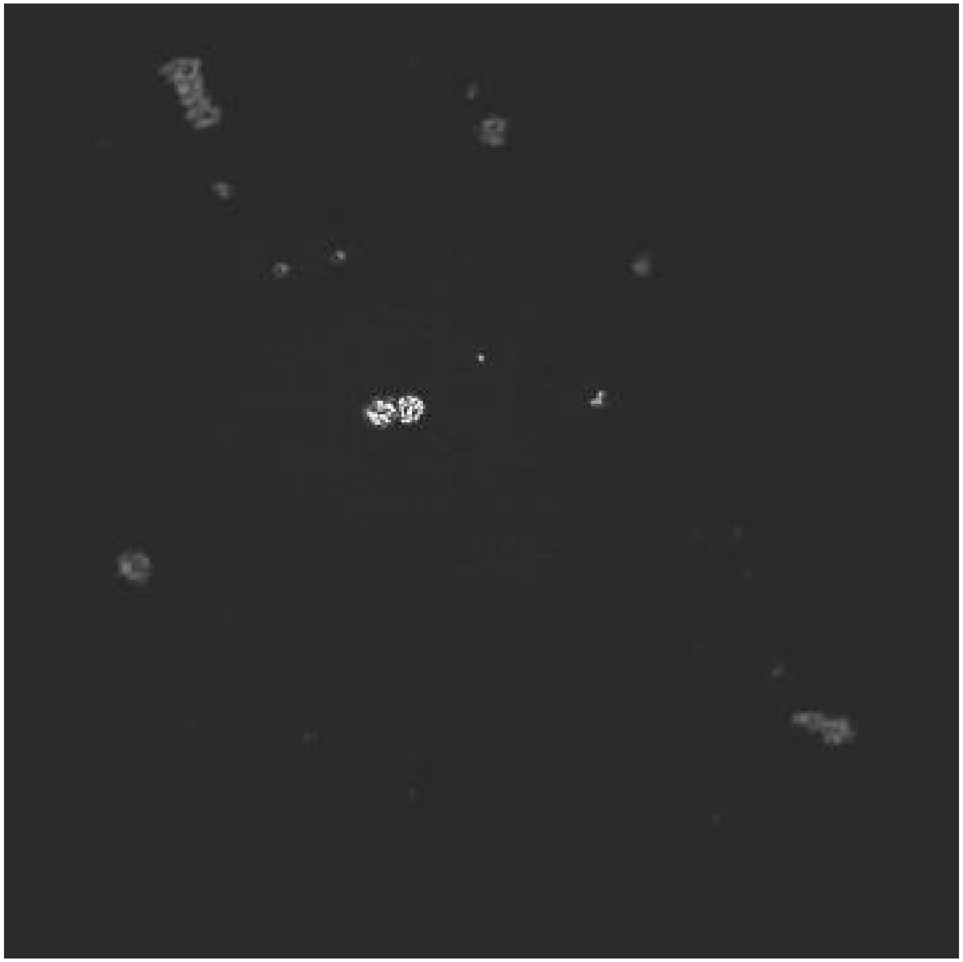
No use of a separate image to find low frequency intensities which were left to self adjust

Support was determined by the SEM image and was a set of 3 circles drawn around each cluster

Zero order self adjusts to the average level needed

Si frame has wedge angle of 54° and 1/e intensity attenuation at 600 eV of 0.6 micron

RECONSTRUCTION OF A WIDER FIELD



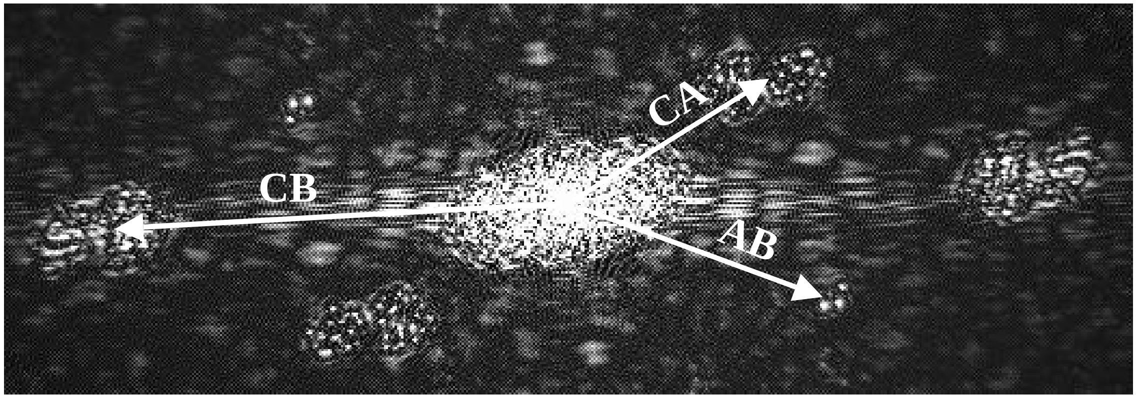
HiO reconstruction of a wider field of view

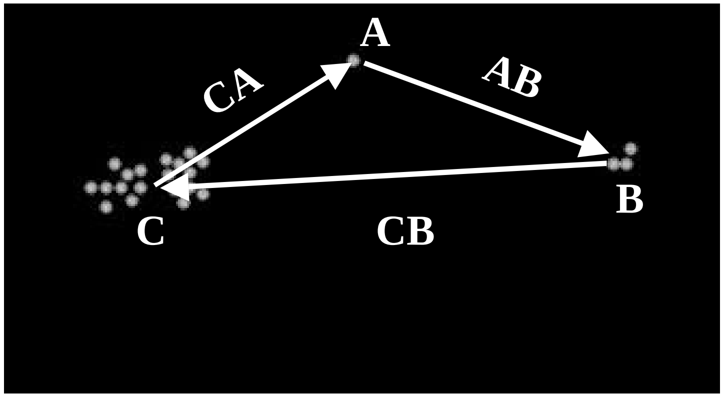
More balls participate due to partial transparency of the window frame

It is appropriate that they should be darker due to absorption in the frame

OBJECT AUTOCORRELATION FUNCTION

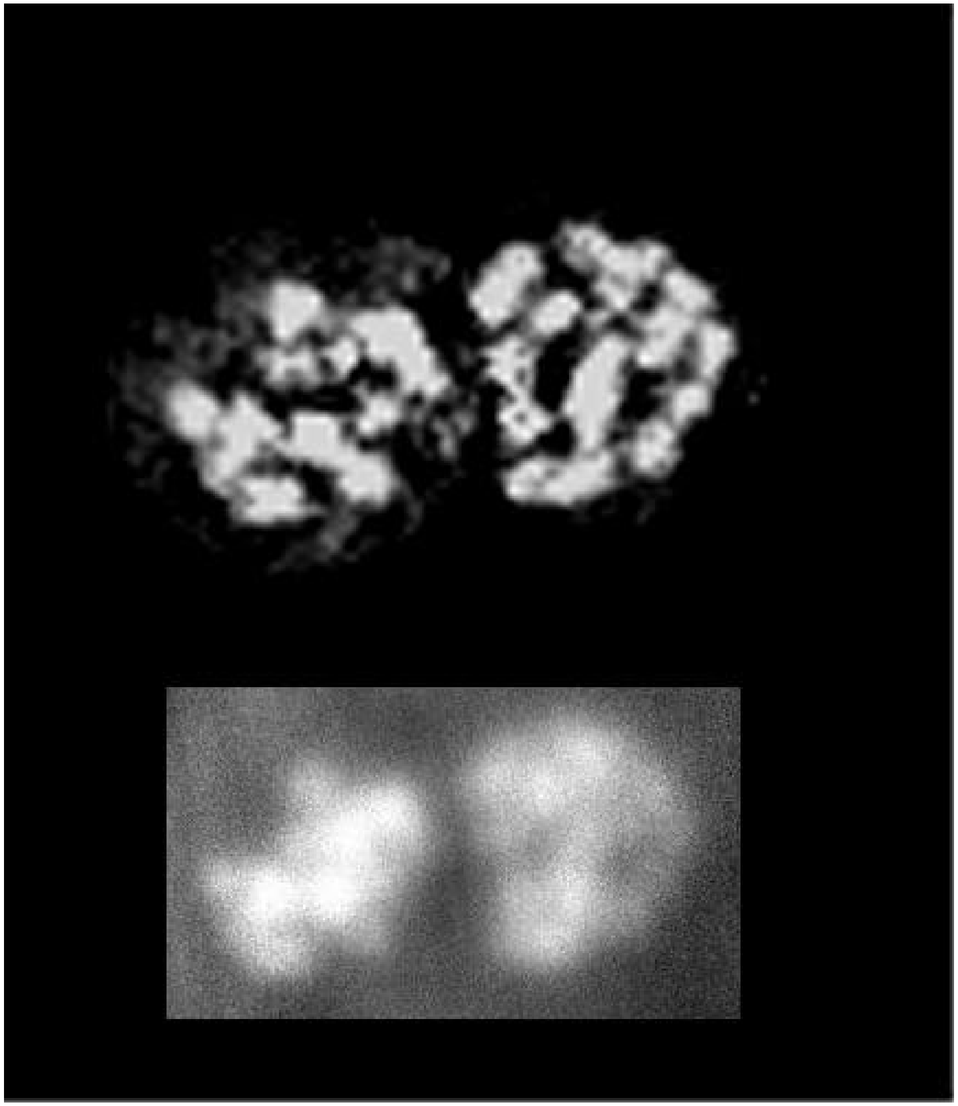
- Autocorrelation of the object found by taking the Fourier transform of the measured diffraction pattern
- Shows the role of separation of parts in determining the support from the autocorrelation
- Since element A is a single ball we see images and twin images of the other two elements formed by Fourier transform holography with a resolution of one ball size (50 nm in this case)





Sketch of the object to explain the features of the autocorrelation

X-RAY-TO-SEM COMPARISON



- X-ray (top) and SEM (bottom) images of the big cluster of balls (SEM taken last)
- The SEM has lower resolution due to charging (effect of the thin-window mounting)
- For the same reason we believe there was some movement of balls *within* clusters but not of one cluster with respect to another
- Such movements may affect the agreement of the two images which is only qualitative for the moment

CONCLUSIONS

- **Developed a beam line and end station for simple diffraction experiments**
- **Measured diffraction patterns of test objects made of gold balls**
- **Tried to address the missing-cone problem by**
  - **Direct methods - not yet successful**
  - **Small windows - very successful**
- **Made a successful reconstruction by using a support derived from the SEM image**
- **Demonstrated interesting properties of the autocorrelation function (the FT of the measured pattern) of an object with separated parts**

APPLICATION OF CRYSTALLOGRAPHIC DIRECT METHODS TO IMAGING

Giacovazzo 1989, Wolfson, 1995, 1997, Sayre 1952

Basic ideas:

- Particular types of sums of structure factors have quite predictable values, e. g. triplets from zero-sum reciprocal lattice vectors tend to add to zero - follows in part from Sayre's equation and its consequence the "tangent formula"
- The method is embodied in standard computer codes - we have used SIR 97 by Giacovazzo et al
- Requirements are:
  - The object consists of atoms with known scattering factors (atomicity)
  - The data extends to atomic resolution - we can do this because our atoms are gold balls!
  - There must be about 40 beams per atom of which at least 8 must be strong - we can do this too because (in the absence of periodicity to discretize the pattern) we can regard each pixel as a beam - with a 2-µm window we have a maximum of 1600 50-nm balls and 1024×1024 pixels - thus plenty of beams
- For λ=2.5 nm say and 25 µm CCD pixels at 100 mm distance, the smallest recordable deviation angle (twice the Bragg angle) is 0.25 mr. This is our [1, 0] beam in 2D. The largest periodicity is thus 10 µm. The smallest periodicity (our [512, 0] beam) corresponds to 10 µm/512=20 nm - so resolution limit in this geometry is 10 nm
- Nonrequirements: missing cone no problem, lack of isolation no problem